

# Sandalwood Research Newsletter

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## Biological and Physiological aspects of the *Santalum acuminatum* (Quandong) and its hosts in south Western Australia

by Kushan U Tennakoon and John S Pate

Kushan Tennakoon, a Sri Lankan national, recently completed his Ph.D at the Department of Botany, University of Western Australia (UWA) whilst on an Overseas Postgraduate Research Scholarship and an UWA Research Studentship. Kushan's address after June 1997: Department of Botany, University of Peradeniya, Peradeniya, Sri-Lanka

Ph.D thesis: Biology and physiology of south West Australian woody angiosperm hemiparasites and their hosts. Supervisor: Prof. JS Pate. John S Pate is a Professor at the Department of Botany, UWA, Perth, Western Australia.

Our study examined a range of biological and functional aspects of the quandong (*Santalum acuminatum*) and its principal hosts in pristine bush land of the northern coastal sand plains of south Western Australia. Preliminary studies across the study region showed that quandong spreads clonally by prolific root suckering. Resprouting after fire occurred from bases of burnt shoots and from new suckers forming at intervals along the lateral root stocks of a parent clone. Systemic death of whole clones was observed after particularly intense fire. Recruitment from seed occurred very rarely and mortality rates of seedlings in nature were high.

Assessments based on densities of above ground biomass, ground cover and frequencies of haustoria on host roots indicated that woody N<sub>2</sub> fixers (legumes and *Allocasuarina*) were principal hosts for *S. acuminatum*. Furthermore,  $\delta^{15}\text{N}$  values for shoot dry matter of quandong in study sites lay closest to those of major N<sub>2</sub> fixing hosts suggesting that the parasite was strongly (70% or more) dependent for N from these hosts.

Formation and functional life of

haustoria were closely coordinated with seasonal growth of hosts, with haustoria surviving summer and overlapping functionally with new ones establishing in the next autumn. This strategy assures effective continuity with hosts in terms of abstraction of water and nutrients across and between seasons. Structural studies of haustoria show the interface with host xylem to be almost entirely comprised of parenchymatous tissue. No luminal continuities were observed between xylem conducting elements of the partners.

Day time transpiration rates and photosynthetic rates of *S. acuminatum* were consistently less and water use efficiencies closely similar to those of the principal host *Acacia rostellifera*. This suggested that quandong might depend heavily on its hosts for carbon. Subsequent studies examining xylem sap composition of host and parasite provided evidence that *S. acuminatum* would gain nearly one third of its

carbon requirements heterotrophically from uptake of amino acids, organic acids and sugars from xylem of its hosts. Gas exchange data obtained for *S. acuminatum* and *A. rostellifera* at a series of intervals in each season of a year correlated well with carbon isotope discrimination values ( $\delta^{13}\text{C}$ ) for dry matter of the canopy leaves of the same two species. The specific hydraulic conductivity values for water flow through roots of quandong were consistently lower than those recorded for *Acacia*. This was viewed as evidence of conservatism in water use by the parasite, just as suggested above from the gas exchange studies.

Solute composition of root xylem sap of common native hosts of quandong was compared with that of corresponding xylem sap and ethanolic extracts of endophytic tissues of haustoria of the hemiparasite. The data (Fig. 1) indicated only limited direct flow of amino compounds between xylem streams of hosts and parasite, a finding consistent with the above mentioned absence of lumen-to-lumen xylem vessel continuity between host and parasite. Proline predominated in the haustorium and xylem of quandong, but was at negligible levels in xylem of most hosts. Sucrose, fructose, glucose malate and citrate were at high levels in all saps, and fructose was especially prominent in *Santalum*. Infiltration of xylem of haustoria-bearing root segments of major host (*A. rostellifera*) with a range  $^{15}\text{N}$  labeled substrates resulted in 40-80% of the  $^{15}\text{N}$  of endophytes of the attached haustoria being recovered as proline. The study concluded that haustoria

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of *Santalum* function as major sites of synthesis and export of proline and might therefore play an important role in osmotic adjustment of quandong in relation to acquisition of water from hosts under differing levels of stress.

While *S. acuminatum* has so far proved of no significance to the sandalwood industry, it is hoped that approaches employed and findings gained in the present study provide

insight on how other species of the genus might behave in natural or plantation situations.

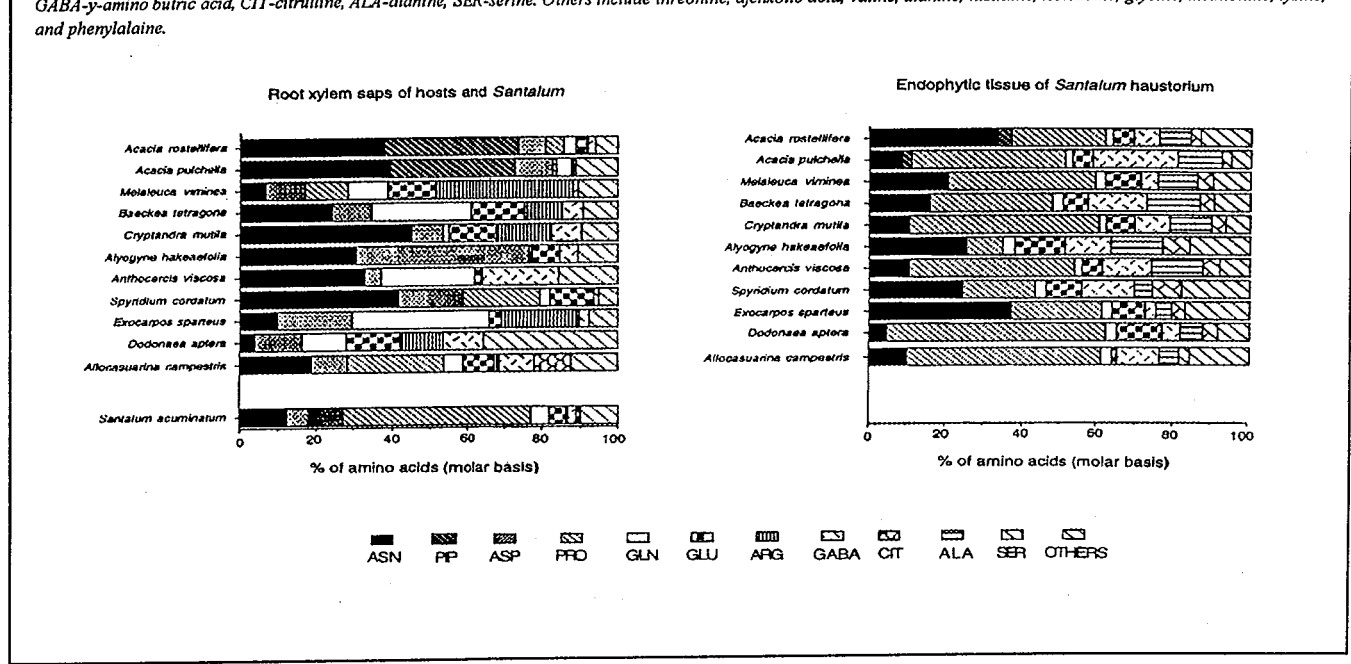
The collaboration of Professor George Stewart and Mr. David Arthur in this study is gratefully acknowledged.

The work summarized above has been accepted to be published in "Annals of Botany" as a series of two papers:

Tennakoon KU, Pate JS, Arthur D. 1997. Ecophysiological aspects of the woody root hemiparasite *Santalum acuminatum* (R. Br.) A. DC. and its common hosts in south Western Australia. *Annals of Botany*. In press.

Tennakoon KU, Pate JS, Stewart GR. 1997. Haustorium-related uptake and metabolism of xylem solutes by the root hemiparasitic shrub *Santalum acuminatum* (R. Br.) A. DC. (Santalaceae). *Annals of Botany*. In press.

Figure 1: The percentage composition (molar basis) of amino acids in root xylem sap of *Santalum acuminatum* and of a range of commonly parasitized host species and in ethanolic extracts of endophytic tissue of haustoria formed by the parasite on the same host species. Note that amino acid pools of haustorial tissue show noticeable differences from those of respective host xylem saps. Code for amino compounds: ASN-asparagine, PIP-pipecolic acid, ASP-aspartic acid, PRO-proline, GLN-glutamic acid, ARG-arginine, GABA-y-amino butric acid, CIT-citrulline, ALA-alanine, SER-serine. Others include threonine, djenkolic acid, valine, alanine, histidine, isoleucine, glycine, methionine, lysine, and phenylalanine.



## Direct Somatic Embryogenesis from Mature Embryos of Sandalwood

by Ravishankar Rai and Jenny McComb

Dr Ravishankar Rai conducted this research whilst on a UNESCO short-term Fellowship in biotechnology at Murdoch University, Perth, Western Australia. Dr Rai's current address: Department of Botany, University of Mysore, Manasagangotri, Mysore, India, 570 006. Jen McComb is an Associate Professor at the Division of Sciences, Biological Sciences, Murdoch University, Perth, Western Australia.

Indian sandalwood (*Santalum album* L.) is closely linked to the culture and religion of many Asian countries and has been valued for its perfumed wood and oils for thousands of years.

Somatic embryogenesis has many potential advantages for mass

propagation and genetic improvement of hardwood forest trees. Somatic embryogenesis has been achieved in a number of angiosperms, but success has been limited with woody plants. Indian sandalwood embryoids have been regenerated from callus of somatic embryos, endosperm and

stem segments by Lakshmi Sita, Rao and Rangaswamy. This report describes the induction of somatic embryos directly on mature zygotic embryo and their regeneration potential.

The seeds of sandalwood were disinfected and cultured on

Murashige and Skoong (MS) medium supplemented either with coconut milk (20% v/v) and casein hydrolysate or with kinetin (1mg/1ml). After ten days on germination medium, embryos were aseptically removed and planted on the same germination medium, or on MS with different concentrations of cytokinins. All cultures were maintained  $25 \pm 2^{\circ}\text{C}$  and 16h photoperiod.

Contrary to earlier observations, 95 per cent of excised embryos cultured with kinetin (1mg/l) showed normal germination and developed into normal seedlings within four to five weeks of culture.

Zygotic embryos cultured on MS medium with BAP (2mg/l) developed a cluster of globular embryos on their surface without an intervening callus phase. The globular embryos continued to develop through the heat and torpedo stages on the same medium resulting in cotyledon-stage embryos. Many embryos formed in vitro showed the normal features characteristic of seed embryos, but some were atypical and abnormal.

When the primary somatic embryoids derived from mature sexual embryos were subcultured on a medium with a low concentration of BAP, secondary somatic embryoids arose directly from the surface of the primary embryoids. Secondary embryogenesis has great potential for mass propagation and repetitive embryogenesis and can also be used for genetic transformation.

To promote maturation and germination, somatic embryos were transferred to germination medium (half-strength MS with 0.5mg/l Indole-acetic acid) and incubated under the same culture conditions as noted earlier.

This report is the first on in vitro regeneration via direct, rather than indirect somatic embryogenesis of sandalwood.

## Workshop on Sandalwood in Papua New Guinea

by Derek Bosimbi

*Derek Bosimbi is the Manager of the PNGFA National Tree Seed Centre, Bulolo, Morobe Province*

From 8 - 14 November 1996 a short but intensive and informative workshop on Sandalwood was conducted in Port Moresby for officers of the National Forest Service. Among other things, all aspects of sandalwood propagation from seed collection to nursery production and plantation establishment were covered. A resource person from Australia, sponsored by the Australian Centre for International Agricultural Research (ACIAR) and the Department of Conservation and Land Management (CALM), conducted the workshop.

Since there was only limited information on sandalwood in the country, especially the local species, *Santalum macgregorli*, the workshop was a 'springboard' for renewed interest and activities to ensure that this low volume high value species does not disappear as a result of over exploitation.

After an intensive introduction to nursery and plantation techniques, including pot hosts, potting medium, intermediate and plantation host compatibility, it was unanimously agreed that further and immediate work was needed. A sandalwood strategy was developed during the workshop's closing session identifying priorities and action items. The document was entitled 'Strategy for the Research, Development and Management of Sandalwood in PNG'.

Following the development of the strategy document a programme schedule was compiled. The main points for consideration were (i) research to include collection of past and current literature on sandalwood, (ii) identification of various seed sources, (iii) local village sandalwood awareness program and (iv) market

exploration. Local village awareness and seed collection was to be implemented immediately.

Programme activities identified during the workshop have already commenced, including a seed purchase trip to Iokea in the Gulf Province. Seed collected from this trip has been passed onto the Forest Research Institute in Lae for germination testing and nursery research.

Overall the workshop was rewarding as it highlighted the strong interest in sandalwood and brought awareness to the PNGFA of our native species. On behalf of the workshop participants I would like to thank ACIAR and CALM for funding the Australian resources persons' visit to PNG.

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Articles on a range of *Santalum* species research and management issues are welcomed by the Sandalwood Research Newsletter. If you wish to contribute an article to the SRN or wish to be included on the SRN mailing list please write to the Editor stating your name, organisation and postal address.

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INTERNATIONAL SEMINAR ON SANDALWOOD AND ITS PRODUCTS  
BANGALORE, INDIA.

Probable date: 18 - 19 December 1997

Venue: Institute of Wood Science and Technology, Bangalore, India

Organised by: Indian Council of Forestry Research and Education and  
Karnataka State Forest Department, Bangalore

**Introduction**

*Santalum* species are distributed widely, from India, Indonesia, throughout much of Australia, Papua New Guinea and many Pacific island countries, as far east as the Juan Fernandez Islands near the coast of South America. The fragrant heartwood of these species, commonly known as sandalwood, is of high commercial significance to many rural based economies. With very few exceptions sandalwood has endured a level of exploitation almost unrivalled by other timbers.

A resurgence in *Santalum* species research and management has generated interest in sandalwood as a potential high value plantation crop in many countries. A South Pacific sandalwood seminar hosted by CIRAD-Foret, New Caledonia, in August 1994 brought together participants from eleven countries involved in sandalwood research and management within the South Pacific region. Following the success of the South Pacific sandalwood seminar the Indian Council of Forestry Research and Education (ICFRE) and the Karnataka State Forest Department (KSFD) are proposing to host an international sandalwood seminar in December 1997.

India has a rich sandalwood heritage. It has been the 'corner-stone' of the global sandalwood industry for 20 - 30 centuries. India has a sound research base in many aspects of *S. album* plantation silviculture, tree improvement, chemistry and utilisation. Mainly, through research undertaken at the Institute of Wood Science and Technology (IWST) and the former Sandalwood Research Centre. A monograph on sandalwood was published by the IWST in 1992. Since then considerable research has been undertaken on sandalwood silviculture, genetics and pest and diseases. As a result, ICFRE and KSFD are proposing to host an international seminar to facilitate the dissemination of information from countries and organisations engaged in sandalwood research and management.

**Scope:** The seminar aims to generate awareness of the global importance of *Santalum* species and to further the cause of sandalwood research, development, sustainable utilisation and conservation. The seminar will have preliminary lectures, contributory papers and discussions on :

1. Silviculture and Management
2. Chemistry and Utilisation
3. Pest and Diseases and their management
4. Role of *Santalum* species in farm forestry
5. International sandalwood Regulations, Laws and Acts.

ICFRE and KSFD welcomes international participants to give paper contributions, including general update papers on their country's involvement in sandalwood.

**Language:** English is the official language for the seminar.

Contact Persons: Sri. H. S. Ananthapadmanbha  
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E-Mail: IWST.MIS.@X400.NICGW.NIC-IN

Further information on the seminar will be circulated on receipt of the completed preliminary registration form.

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Preliminary registration form.

Name: ..... Company/Organisation: .....

Street/PO: ..... City: ..... State: .....

Zip/Post Code: ..... Country: .....

Phone: ..... Fax: ..... E-Mail: .....

Yes, I want to give a presentation.

Title: .....

Yes, I want to be included onto the conference mailing list. Only respondents will receive future correspondence

Return to: H.S. Ananthapadmanbha/C. R. Rangaswamy, Institute of Wood Science and Technology, Malleswaram, Bangalore - 560 003. INDIA.